CLAIMS

1. A method for preparing one or more pleuromutilins comprising the steps of:
a) culturing a pleuromutilins-producing microorganism in a liquid culture medium; and
b) extracting the pleuromutilins from the unfiltered culture medium with a water immiscible organic solvent.

2. A method for preparing one or more pleuromutilins comprising the steps of:
a) culturing a pleuromutilins-producing microorganism in a liquid culture medium;
b) extracting pleuromutilins from the unfiltered culture medium with a water immiscible organic solvent;
c) concentrating the extracted pleuromutilins; and
d) crystallising the pleuromutilins.

3. A method according to claim 2 wherein the extracted pleuromutilins (Step b) or the concentrated pleuromutilins (Step c) are decolourised using activated carbon.

4. A method according to any one of the preceding claims for preparing pleuromutilin.

5. A method according to any one of the preceding claims wherein the pleuromutilins-producing microorganism is a *Clitopilus* species, an *Octojuga* species, a *Gerronema* species, a *Psathyrella* species, or a mutant thereof.

6. A method according to claim 5 wherein the pleuromutilins-producing microorganism is *Clitopilus passeckerianus* NRRL 3100/DSM 1602, *Clitopilus passeckerianus* CBS 299.35, *Clitopilus passeckerianus* CBS 330.85, *Clitopilus pinsitus* CBS 623.70, *Clitopilus hobsonii* CBS 270.36, *Octojuga pseudopinsitus* NRRL11179, *Gerronema josserandii* CBS 309.36, *Psathyrella subatrata* CBS 325.39, or a mutant thereof.

7. A method according to claim 6 wherein the pleuromutilins-producing microorganism is *Clitopilus passeckerianus* NRRL 3100 or a mutant thereof

8. A method according to any one of the preceding claims wherein the water immiscible organic solvent is an aromatic hydrocarbon or a water immiscible aliphatic ketone.

9. A method according to claim 8 wherein the aromatic hydrocarbon is toluene.

10. A method according to claim 8 wherein the water immiscible aliphatic ketone is MIBK.

11. A method according to any one of the preceding claims wherein the extraction is conducted at about 10°C to about 50°C.

12. A method according to any one of the preceding claims wherein the pH of the aqueous solution prior to extraction is in the range pH 6 to 8.

13. A method according to any one of the preceding claims wherein a ratio of 4:1 to 1:4 equivalent volume of organic solvent to unfiltered culture medium is used for the extraction.

14. A method according to any one claims 2 to 13 wherein the pleuromutilins are directly crystallised from toluene or MIBK.

15. A method according to claim 14 wherein the pleuromutilins are directly crystallised from toluene and the concentration of the toluene solution used for crystallisation is from 10% to 50% w/w.

16. A method according to claim 14 or 15 wherein the pleuromutilins are directly crystallised from toluene and the initial temperature of the toluene is from 60°C to 70°C, followed by cooling to from 0°C to 5°C for 8-10 hours.

17. A method according to claim 14 wherein the pleuromutilins are directly crystallised from MIBK and the concentration of the MIBK solution used for crystallisation is from 20% to 45% w/w.

18. A method according to claim 14 or 15 wherein the pleuromutilins are directly crystallised from MIBK and the initial temperature of the MIBK is from 45°C to 60°C, followed by cooling to from 25°C to 35°C.

19. A method according to any one of claims 2 to 13 wherein the pleuromutilins are directly crystallised from MIBK and a miscible non-polar solvent.

20. A method according to claim 19 wherein the miscible non-polar solvent is heptane.

21. A method according to any one of claims 2 to 20 wherein the crystallised pleuromutilins are further purified by recrystallisation.

22. A method according to claim 21 wherein mutilin 14-acetate is selectively removed from the crystallised pleuromutilins by recrystallisation with ethyl acetate and heptane.

23. A method according to claim 21 or claim 22 wherein the concentration of pleuromutilins used for recrystallisation is from 20% to 40% w/w.

24. A method according to any one of claims 21 to 23 wherein the initial temperature is from 45 °C to 50 °C, followed by cooling to from 15°C to 25 °C.

25. A method according to claim 24 followed by heptane addition and further cooling to 0 °C to 5 °C.

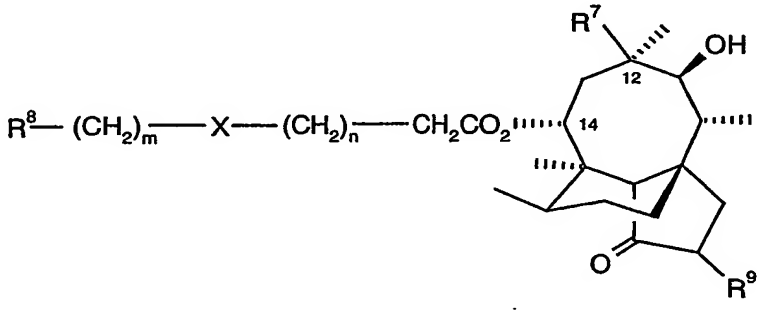
26. A method according to claim 21 wherein mutilin 14-acetate is selectively removed from the crystallised pleuromutilins by recrystallisation with MIBK and heptane.

27. A method according to claim 21 or claim 26 wherein the concentration of pleuromutilins used for recrystallisation is from 20% to 45% w/w.

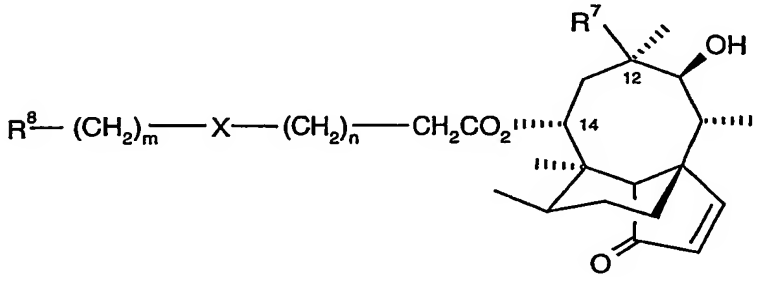
28. A method according to any one of claims 21, 26 and 27 wherein the initial temperature is from 45 °C to 65 °C.

29. A method of preparing a semi-synthetic pleuromutilins derivative comprising preparation of pleuromutilins by a process claimed in any one of the preceding claims.

30. A method according to claim 29 wherein the semi-synthetic pleuromutilins derivative is a compound of general formula (4A) or (4B):



(4A)



(4B)

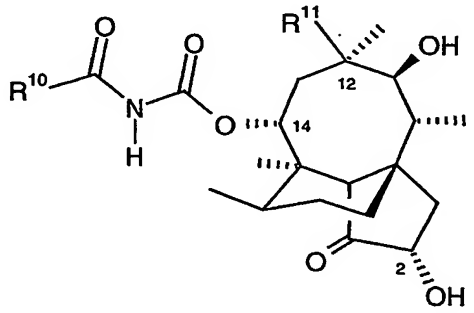
in which:
each of n and m is independently 0, 1 or 2;

X is selected from -O-, -S-, -S(O)-, -SO_2-, -CO.O-, -NH-, -CONH-, -NHCONH- and a bond;
R^7 is vinyl or ethyl;
R^8 is an optionally substituted non-aromatic monocyclic or bicyclic group containing one or two basic nitrogen atoms and attached through a ring carbon atom;
R^9 is H or OH; or
the moiety $R^8(CH_2)_mX(CH_2)_nCH_2COO$ at position 14 of (4A) or (4B) is replaced by $R^aR^bC{=}CHCOO$ in which one of R^a and R^b is hydrogen and the other is R^8 or R^a and R^b together form R^8; or
a pharmaceutically acceptable salt thereof.

31. A method according to claim 30 wherein the semi-synthetic pleuromutilins derivative is a compound of formula (4A) or (4B) wherein R^8 is selected from optionally substituted piperidinyl, pyrrolidyl, quinuclidinyl, azabicyclo[2.2.1]heptyl, azabicyclo[4,3,0]nonyl, azabicyclo[3.2.1]octyl, azabicyclo[3,3,0]octyl, azabicyclo[2.2.2]octyl, azabicyclo[3.2.1]octenyl, azabicyclo[3.3.1]nonyl and azabicyclo[4.4.0]decyl.

32. A method according to claim 30 or 31 wherein the semi-synthetic pleuromutilins derivative is a compound of formula (4A) or (4B) wherein R^8 is substituted by alkyl, alkyloxy, alkenyl or alkenyloxy, which are optionally further substituted by one or more groups selected from aryl, heterocyclyl, (C_{1-6})alkoxy, (C_{1-6})alkylthio, aryl(C_{1-6})alkoxy, aryl(C_{1-6})alkylthio, amino, mono- or di-(C_{1-6})alkylamino, cycloalkyl, cycloalkenyl, carboxy and esters thereof, amides of carboxy, ureido, carbamimidoyl (amidino), guanidino, alkyl-sulfonyl, amino-sulfonyl (C_{1-6})acyloxy, (C_{1-6})acylamino, azido, hydroxy, and halogen.

33. A method according to claim 29 wherein the semi-synthetic pleuromutilins derivative is a compound of general formula (5):



(5)

in which:
R^{10} is a 5- or 6-membered optionally substituted heteroaryl group; and
R^{11} is vinyl or ethyl;

or a pharmaceutically acceptable salt thereof.